11

6502985446

PATENT Atty. Docket No: 0113.410US USSN 09/373.333

Courtesy Copy Of Pending Claims For

USSN 09/373,333 As Amended

- 4. (thrice amended) A method of obtaining a recombinant herbicide tolerance nucleic acid that encodes an herbicide tolerance polypeptide, wherein expression of the herbicide tolerance polypeptide at effective levels in a cell renders the cell tolerant towards an herbicide, the method comprising:
- providing a plurality of nucleic acid segments derived from a (a) plurality of variant forms of a gene, wherein the gene encodes a UDP-N-acetylglucosamine enolpyruvyltransferase;
- recombining the plurality of nucleic acid segments to produce a library of recombinant nucleic acids;
- (c) screening the library to detect a recombinant herbicide tolerance nucleic acid that encodes an herbicide tolerance polypeptide that catalyzes the conversion of phosphoenolpyruvate plus shikimate-3-phosphate to 5-enolpyruvylshikimate-3-phosphate, wherein expression of the herbicide tolerance polypeptide at effective levels in the cell renders the cell tolerant towards the herbicide; and
- recovering the recombinant herbicide tolerance nucleic acid that (d) encodes an herbicide tolerance polypeptide having EPSP synthase activity.
- 6. (twice amended) The method of claim 4, wherein the plurality of variant forms comprises allelic or interspecific variants of the gene.
- 7. (twice amended) The method of claim 4, wherein the plurality of variant forms is produced by synthesizing a plurality of nucleic acids homologous to the gene.
- 8. (twice amended) The method of claim 4, wherein the plurality of variant forms is produced by error-prone transcription of the gene or by replication of the gene in a mutator cell strain.
 - 11. (amended) The method of claim 4, wherein the herbicide is glyphosate.
- 14. (twice amended) The method of claim 4, wherein the library of recombinant nucleic acids is present in a population of cells.
- 15. (as filed) The method of claim 14, wherein the screening comprises growing the population of cells in or on a medium comprising the herbicide and detecting a physical difference between the herbicide and a modified form of the herbicide produced by the cells.

П

- 16. (as filed) The method of claim 15, wherein the physical difference between the herbicide and the modified form of the herbicide is detected by a difference in fluorescence or absorbance between the herbicide and the modified form of the herbicide.
- 18. (as filed) The method of claim 14, wherein the screening comprises growing the population of cells in or on a medium comprising the herbicide and selecting for enhanced growth of the cells in the presence of the herbicide.
- 19. (amended) The method of claim 18, wherein enhanced growth of the cell requires the expression of the herbicide tolerance polypeptide at effective levels in the cell.
- 20. (amended) The method of claim 19, wherein enhanced growth of the cell requires a product of a reaction catalyzed by the herbicide tolerance polypeptide.
- 22. (amended) The method of claim 19, wherein the cells are an AroA strain of bacteria, the herbicide is glyphosate, and the recombinant herbicide tolerance nucleic acid encodes a polypeptide that catalyzes the conversion of phosphoenolpyruvate plus shikimate-3-phosphate to 5-enolpyruvylshikimate-3-phosphate.
- 23. (twice amended) The method of claim 4, the method further comprising screening the library for an additional activity that confers tolerance to an additional herbicide.
- 24. (twice amended) The method of claim 4, wherein the recombining is performed in a population of cells.
- 28. (twice amended) The method of claim 4, wherein the method further comprises:
- (d) recombining a recombinant herbicide tolerance nucleic acid detected in step (c) with an additional nucleic acid, wherein the additional nucleic acid is the same or different from the plurality of variant forms of a gene of step (a), to produce an additional library of recombinant nucleic acids; and
- (e) screening the additional library to detect a recombinant herbicide tolerance nucleic acid that encodes an herbicide tolerance polypeptide, wherein expression of the herbicide tolerance polypeptide at effective levels in the cell renders the cell tolerant towards the herbicide; and, optionally

repeating steps (d) and (e).

30. (twice amended) The method of claim 4, wherein the library of recombinant nucleic acids is present in bacterial cells and the screening step comprises:

pooling a plurality of cells each comprising a separate member of the library produced in step (b);

screening the resulting pooled cells to detect a recombinant herbicide tolerance nucleic acid that encodes an herbicide tolerance polypeptide, wherein expression of the herbicide tolerance polypeptide at effective levels in the cell renders the cell tolerant towards the herbicide; and

isolating the recombinant herbicide tolerance nucleic acid.

- 32. (twice amended) The method of claim 4, further comprising transducing the recombinant herbicide tolerance nucleic acid into a plant.
- 33. (twice amended) The method of claim 4, further comprising transducing the recombinant herbicide tolerance nucleic acid into a plant and testing the resulting transduced plant for tolerance to an herbicide.
- 34. (twice amended) The method of claim 4, further comprising transducing the recombinant herbicide tolerance nucleic acid into a plant and breeding the plant with another plant strain of the same species, and selecting resultant offspring for tolerance to an herbicide.
- 35. (twice amended) A library of recombinant nucleic acids made by the method of claim 4.
- 36. (as filed) The library of claim 35, wherein the library is a phage display library.
- 37. (twice amended) A recombinant herbicide tolerance nucleic acid made by the method of claim 4.
- 61. (amended) The method of claim 4, further comprising isolating or recovering the recombinant herbicide tolerance nucleic acid.
 - 62. (as filed The method of claim 4, wherein the gene is a MurA gene.
 - 63. (as filed) The method of claim 62, wherein the gene is a bacterial MurA gene.
- 64. (as filed) The method of claim 63, wherein the bacterial *MurA* gene is selected from the group consisting of the genes corresponding to GenBank Accession Numbers M76452, Z11835, AF142781 and X96711.
 - 65. (amended) The method of claim 4, wherein:
- step (a) further comprises providing an EPSP synthase nucleic acid segment derived from a gene that encodes an EPSP synthase; and
- step (b) further comprises recombining the EPSP synthase nucleic acid segment with the plurality of nucleic acid segments to produce the library of recombinant nucleic acids.

FROM-MAXYGEN

PATENT Atty. Docket No: 0113.410US USSN 09/373,333

66. (as filed) The method of claim 65, wherein the EPSP synthase nucleic acid segment is derived from the S3P binding region of an EPSP synthase gene.

67. (as filed) The method of claim 28, wherein the additional nucleic acid is derived from a gene that encodes an EPSP synthase.